

SEQUENCE LISTING

<110> INSTITUT PASTEUR
CENTRE NATIONAL DE LA RECHERCHE SCIENTIFIQUE

<120> Method for in vivo modification of the synthesis activity of a metabolite by modification of a gene the activity of which is not the original activity.

<130> BIF 023274 PCT

<140> PCT/US/FR03/xxxxx
<141> 2003-03-28

<150> FR 03 03910
<151> 2003-03-28

<160> 15

<170> Patentln version 3.1

<210> 1
<211> 474
<212> DNA
<213> Lactobacillus leichmannii

<220>
<221> misc_feature
<222> (1). (474)
<223> Coding region of the N-deoxyribosyltransferase gene (dtp)

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<400> 1
atgccaaaaa agacgatcta cttcgggtgcc ggctggttca ctgaccgcca aaacaaagcc      60
tacaaggaag ccatggaagc cctcaaggaa aacccaacga ttgacctgga aaacagctac      120
gttcccctgg acaaccagta caagggatc cgggttgatg aacaccgga atacctgcat      180
gacaaggttt gggctacggc cacctacaac aacgacttga acgggatcaa gaccaacgac      240
atcatgctgg gtgtctacat ccctgacgaa gaagacgtcg gcctgggcat ggaactgggt      300
tacgccttga gccaaaggcaa gtacgtcctt ttggtcatcc cggacgaaga ctacggcaag      360
ccgatcaacc tcatgagctg gggcgtcagc gacaacgtga tcaagatgag ccagctgaag      420
gacttcaact tcaacaagcc gcgcttcgac ttctacgaag gtgccgtata ctaa          474
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<210> 2
<211> 157
<212> PRT
<213> Lactobacillus leichmannii

<220>
<221> MISC_FEATURE
<222> (1)-(157)

<223> N-deoxyribosyltransferase carrying the mutation G9S.

<220>

<221> MISC_FEATURE

<222>(9).._(9)

<223> serine/glycine mutation

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Met Pro Lys Lys Thr Ile Tyr Phe Ser Ala Gly Trp Phe Thr Asp Arg
1           5           10           15

Gln Asn Lys Ala Tyr Lys Glu Ala Met Glu Ala Leu Lys Glu Asn Pro
          20           25           30

Thr Ile Asp Leu Glu Asn Ser Tyr Val Pro Leu Asp Asn Gln Tyr Lys
          35           40           45

Gly Ile Art Val Asp Gly His Pro Gly Tyr Leu His Asp Lys Val Trp
          50           55           60

Ala Thr Ala Thr Tyr Asn Asn Asp Leu Asn Gly Ile Lys Thr Asn Asp
65           70           75           80

Ile Met Leu Gly Val Tyr Ile Pro Asp Glu Glu Asp Val Gly Leu Gly
          85           90           95

Met Glu Leu Gly Tyr Ala Leu Ser Gln Gly Lys Tyr Val Leu Leu Val
          100          105          110

Ile Pro Asp Gly Asp Tyr Gly Lys Pro Ile Asn Leu Met Ser Trp Gly
          115          120          125

Val Ser Asp Asn Val Ile Lys Met Ser Gln Leu Lys Asp Phe Asn Phe
          130          135          140

Asn Lys Pro Arg Phe Asp Phe Tyr Glu Gly Ala Val Tyr
145           150           155
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<210> 3

<211> 474

<212> DNA

<213> Lactobacillus leichmannii

<220>

<221> misc_feature

<222> (1)..(474)
 <223> Coding sequence of mutated N-deosyribosyltransferase(NTD*).

<400> 3
 atgccaaaaa agacgatcta cttcagtgcc ggctggttca ctgaccgcca aaacaaagcc 60
 tacaaggaag ccatggaagc cctcaaggaa aaccaaacga ttgacctgga aaacagctac 120
 gttccccttg acaaccagta caagggtatc cgggttgatg aacacccgga atacctgcat 180
 gacaagggtt gggctacggc cacctacaac aacgacttga acgggatcaa gaccaacgac 240
 atcatgctgg gcgtctacat ccctgacgaa gaagacgtcg gcctgggcat ggaactgggt 300
 tacgccttga gccaaaggcaa gtacgtcctt ttggtcatcc cggacgaaga ctacggcaag 360
 ccgatcaacc tcatgagctg gggcgtcagc gacaacgtga tcaagatgag ccagctgaag 420
 gacttcaact tcaacaagcc gcgcttcgac ttctacgaag gtgccgtata ctaa 474

<210> 4
 <211> 32
 <212> DNA
 <213> artificial sequence

<220>
 <221> primer
 <222> (1)..(32)
 <223> primer codBL for the amplification of the PyrC gene

<220>
 <221> misc_feature
 <222> (1)..(1)
 <223> n is a nucleotide comprising a base A, T, C or G.

<220>
 <221> misc_feature
 <222> (2)..(2)
 <223> n is a nucleotide comprising a base A, T, C or G.

<220>
 <221> misc_feature
 <222> (3)..(3)
 <223> n is a nucleotide comprising a base A, T, C or G.

<400> 4

nnnccccggc ttcttgctcg cttctcgttt gg 32

<210> 5
 <211> 29
 <212> DNA

<213> Artificial sequence

<220>

<221> primer

<222> (1) .. (29)

<223> primer cynTR for amplifying the pyrC gene.

<220>

<221> misc_feature

<222> (1).7(1)

<223> n is a nucleotide comprising a base A, T, C or G.

<220>

<221> misc_feature

<222> (2). (2)

<223> n is a nucleotide comprising a base A, T, C or G.

<400> 5

nnggatccgt ttgaccgtag cgggcgaac

29

<210> 6

<211> 29

<212> DNA

<213> Artificial sequence

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<221> primer

<222> (1)..(29)

<223> Primer codBR allowing deletion of the CodA gene for the construction of the PAK9 strain.

<220>

<221> misc_feature

<222> (1)..(29)

<223> n is a nucleotide comprising a base A, T, C or G.

<400> 6

ngaattctta ttcgacactg ttagcctcc

29

<210> 7

<211> 27

<212> DNA

<213> Artificial sequence

<220>

<221> primer

<222> (1)..(27)

<223> Primer cynTL used in order to delete the CodA gene in the construction of the PAK9 strain.

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<221> misc_feature
<222> (1)..(1)
<223> n is a nucleotide comprising a base A, T, C or G.

<400> 7
ngaattcacg actgggttac agcgagc

27

<210> 8
<211> 35
<212> DNA
<213> Artificial sequence

<220>
<221> Primer
<222> (1)..(35)
<223> Primer ycEL used to amplify a DNA fragment of E.coli (M G1655) containing the pyrC gene.

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<222> (1)..(1)
<223> n is a nucleotide comprising a base A, T, C or G.

<220>
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<222> (2)..(2)
<223> n is a nucleotide comprising a base A, T, C or G.

<220>
<221> misc feature
<222> (3)-(3)
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<400> 8
nnncccgggg ccgacctgct ggcccactct gacgg

35

<210> 9
<211> 38
<212> DNA
<213> Lactobacillus leichmannii

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<221> Primer
<222> (1)..(38)
<223> Primer dinR used to amplify a DNA fragment of E.coli (M G1655) containing the pyrC gene.

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<222> (1)..(1)
<223> n is a nucleotide comprising a base A, T, C or G.

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<221> misc_feature
<222> (2)..(2)
<223> n is a nucleotide comprising a base A, T, C or G.

<400> 9
nnggatcccc cggcggcagc gcctacggaa ccgctgcc

38

<210> 10
<211> 37
<212> DNA
<213> *Lacobacillus leichmannii*

<220>
<221> Primer
<222> (1)..(37)
<223> Primer yceR used for the amplification of transforming plasmid DNA during the preparation of the PAK9 strain.

<220>
<221> Primer
<222> (1)..(37)
<223> Primer yceR used for the amplification of transforming plasmid DNA during the preparation of the PAK9 strain.

<220>
<221> misc_feature
<222> (1)..(1)
<223> Nucleotide comprising a base A, T, C or G.

<400> 10
ngaattctta atcagtaaat ggaatgacaa ttctgcc

37

<210> 11
<211> 34
<212> DNA
<213> *Lactobacillus leichmannii*

<220>
<221> Primer
<222> (1)..(34)
<223> Primer dinL used for the amplification of transforming plasmid DNA during the preparation of the PAK9 strain.

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<221> misc_feature
<222> (1)..(1)

<223> Nucleotide comprising a base A, T, C or G.

<400> 11
ngaattcaaa tcgtagcttc ctgttgatcat-tagc

34

<210> 12
<211> 22
<212> DNA
<213> Artificial sequence

<220>
<221> Primer
<222> (1)..(22)
<223> Primer FP23 for the amplification of the ntd gene.

<400> 12
cgccagggtt ttcccagtcg cg

22

<210> 13
<211> 23
<212> DNA
<213> Artificial sequence

<220>
<221> Primer
<222> (1)..(23)
<223> Primer RP23 for the amplification of the ntd gene.

<400> 13
agcggataac aatttcacac agg

23

<210> 14
<211> 30
<212> DNA
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<221> Primer
<222> (1)..(30)
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<400> 14
gatatacata tgccaaaaaa gacgatctac

30

<210> 15
<211> 36
<212> DNA
<213> Artificial sequence

<220>
<221> Primer
<222> (1)..(36)
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<220>
<221> misc_feature
<222> (2)..(2)
<223> n is a nucleotide comprising a base A, T, C or G.

<400> 15
nnggatcctt agtatacggc accttcgtag aagtcg

36